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PRACTICAL METHODS

OF

URINE ANALYSIS

For Chemists and Druggists

WITH NOTES ON THE
COMPOSITION OF THE NORMAL AND ABNORMAL
RENAL SECRETIONS

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THIS book is written by a pharmacist who has had nearly a quarter of a century's daily experience in the practical application of the methods described herein.

PRACTICAL METHODS
OF
URINE ANALYSIS

THE usual composition of a twenty-four hours' collection of normal urine secreted by a man averages as follows :

Quantity	40 to 50 fluid ounces.
Total solids	800 to 1,000 grains.

The total solids consist of:

Urea	350 to 600 grains.
Uric acid.	5 to 15 ,,
Chlorine	50 to 150 ,,
Phosphoric acid	30 to 60 ,,
Sulphuric acid	20 to 60 ,,

These with their bases sodium, potassium, ammonium, calcium, and magnesium, with extractive matters, pigments, and traces of hippuric acid, mucus, and epithelial cells, make up the weight of total solids indicated above. In chemical analysis the proportion of some of the above constituents in normal urine is estimated as follows :

Urea.	2 to 3 per cent.
Uric acid	3 to 7 parts per 1,000.
Phosphates as P_2O_5 . . .	2 to 3 ,, ,, ,,
Sulphates as SO_3 . . .	1·5 to 3 ,, ,, ,,
Chlorides as $NaCl$ . . .	5 to 10 ,, ,, ,,

To properly report on a specimen of urine, the following particulars should first be noted :

TABLE I.

1. Preliminary remarks.	Specimen of urine for analysis. Received from. Address. Particulars of collection. Particulars of quantity.

2. Physical characters (normal and abnormal).
    - Translucency.
    - Consistency.
    - Colour.
    - Odour.
    - Froth.
3. Reaction.
4. Specific gravity and total solids.
5. Deposit.

Collection of urine for analysis.—It often happens that urine passed before breakfast, after the night's rest, is quite free from albumen, while that which is secreted later contains albumen in abundance. For this reason a twenty-four hours' collection is best ; but if this is not available, a mixed morning and evening sample will give a good average result. The urine received for examination should be fresh, as after a few days' standing it decomposes and exhales an offensive odour, and is markedly ammoniacal owing to the conversion of urea into carbonate of ammonium, which renders it alkaline. It also becomes turbid from the deposition of phosphates. Putrid urine contains ammonium sulphide, due to the deoxidation of the sulphates by the organic matter. It sometimes simplifies matters if the patient is directed to pass water in two portions and into separate vessels, so that the first, which should be the smaller portion, may be examined for diseases of the urethra, and the second for diseases of the bladder or kidneys. If it be necessary to obtain urine free from bladder or urethral products, the bladder has to be emptied and washed out, and then the pure renal secretion is allowed to pass drop by drop through a catheter.

Quantity secreted daily.—The quantity of urine secreted and passed daily in health averages fifty fluid ounces, but this amount may vary from thirty to as much as eighty ounces, and still be within sound physiological limits. In some forms of structural kidney disease, five or six pints may be passed in the twenty-four hours, while in diabetes as much as three times this quantity may be passed in the same time. Frequent micturition is a marked symptom in kidney and bladder affections, and healthy kidneys secrete a larger quantity of urine during the day than during the night An increase during the night is suggestive of renal mischief or diabetes, and in some stages of atrophic degeneration of the kidneys the nocturnal rise is very considerable. The quantity of urine secreted per diem is diminished :

1. In cases where the specific gravity of the urine is increased (except diabetes).
2. In fevers.
3. In cholera or diarrhœa with sickness.
4. In dropsy (early stages).
5. By exercise, hot weather, or both.
6. In congestion of the kidney from heart disease.
7. During the course of Bright's disease.
8. During the whole period of some kidney diseases, but towards the close in all varieties.
9. In most hepatic diseases and always in cirrhosis.
10. In mechanical obstruction of the ureter from calculi or tumour.
11. In acute scarlatinal nephritis.
12. In shock from passing the catheter or other operations on the urethra.
13. After a fit of hysteria, etc.

Temporary increase is observed :

1. In cold.
2. In nervous excitement.
3. During a fit of hysteria.
4. In asthmatic attacks.
5. After diuretics, etc.

A persistent increase is observed in :

1. Diabetes mellitus.
2. Diabetes insipidus.
3. Certain stages of some kidney diseases.

Generally speaking, increased specific gravity of urine is accompanied by a diminished quantity, while diminished specific gravity is accompanied by an increased quantity ; but to this rule there are some exceptions.

Translucency

Normal urine is transparent at the time of passing, but on standing a cloud of mucus corpuscles and epithelial scales, detached from the surfaces over which the urine has passed, forms in it, and sinks in about twelve hours. The bladder and urethra supply a roundish or oval epithelial cell; and in women, especially during an attack of leucorrhœa, vaginal cells appear plentifully, and resemble the squamous epithelial cells of the mouth. The mucous membrane of the pelvis and ureter supplies irregular caudate or spindle-shaped cells, the presence of which was at one time considered characteristic of cancer. This subject will be dealt with later. With the exception of the muco-epithelial cloud just mentioned, all constituents of normal urine should be in solution. It is only when pathological changes occur in the system that some of these constituents are produced in an abnormal quantity and deposited in an insoluble state, either at the time the urine is secreted or after it has been passed. Thus in gout and fevers, and concentration of urine from other causes, urates are thrown out of the urine on cooling, because more urates are present than the cold urine will retain in solution.

Turbid abnormal urine may arise from :

1. Urates deposited on cooling and which redissolve by heat.
2. Earthy phosphates present on passing, and which do not clear up by heat. The addition of nitric acid clears up earthy phosphates, but not mucus or pus,
3. Pus, muco-pus, or blood.
4. Chyluria, where the fatty globules give the urine a milky appearance, or where a jelly-like fibrinous clot is formed, rising to the surface, forming a white or pinkish jelly, and leaving the urine below turbid. Such urine always contains albumen, but sometimes the appearances are due principally to blood, peptones, and sugar.
5. Bacteria, spermatozoa, or the introduction of foreign matters.
6. Catarrh of the bladder or retention of urine where the ammoniacal products of the decomposition of the urea render any pus present thick and viscid, making the secretion of a tenacious and ropy consistency.
7. Kyestëin, a white film or pellicle formed, after resting a few days, on the surface of urine of pregnant women. This consists of the mould fungus, globules of fat, and crystals of phosphates. The same pellicle has, however, been found in the urine of men, and is therefore not a reliable sign of pregnancy.

Consistency

From what has been said regarding transparent and turbid urine, the consistency can easily be defined as transparent, milky, thickish, semi-fluid, viscid, or ropy.

Colour

Normal urine varies from straw-yellow (*urina potûs*) to golden amber (*urina cibi*), and the abnormal colours may be light yellow, blood-red, reddish to brownish-green, smoky, blue, black, etc. The varieties of abnormally pale urine are generally due to :

1. Administration of tannin (colourless).
2. Copious drinking.
3. Cold weather.
4. A fit of hysteria.
5. Anæmia.
6. Chlorosis.
7. Diabetes.
8. Some forms and stages of kidney diseases.
9. Convalescence from acute diseases.
10. Sex. The urine of women is paler than that of men.

The varieties of abnormally dark urine are generally due to :

1. Drinking little fluid.
2. Concentration of urine.
3. Warm weather.
4. Exercise.
5. Fevers.
6. Diseases of the liver.
7. Addison's disease.
8. Cholera and diarrhœa.
9. Presence of indican, melanin, blood, bile, etc.

Abnormal colour may also be due to medicine, as in the case of tannin, or to the consumption of certain articles of diet.

Carbolic acid, creosote, logwood, and indigo colour the urine dark, or even black.

Santonin colours it a dark bile-like yellow, staining linen.

Rhubarb, senna, and chrysophanic acid give it a red tint.

Odour

In addition to the characteristic aromatic odour of normal urine, certain abnormal odours occur by which the following indications are suggested :

1. Ammoniacal.—Decomposition of urea before or after passing, and the presence of alkali in the urine.
2. New-mown hay.—The saccharine urine of diabetes when passed.
3. Turned milk.—When saccharine urine is fermenting.
4. Chloroform.—Diabetic coma.
5. Sweet briar.—Urine containing much cystin when passed.
6. Sulphuretted hydrogen.—Urine containing much cystin after standing.
7. Sickly odour.—Dyspepsia.
8. Strong odour.—High percentage of urea.
9. Like horse's urine when voided, or shortly after.—Alkalinity due to the administration of a fixed alkali and not due to decomposition of urea.
10. Putrescent odour.—In some diseases of the bladder, kidney, etc.
11. Violet.—Administration of turpentine.
12. Offensive odour.—From decomposition of blood or mucus, or stale normal urine.
13. Copaiba, cubebs, asparagus, garlic, and other medicines or foods give their characteristic odours to the urine.

Froth

The froth of urine quickly disappears in normal urine, but is more or less permanent in albuminous or bilious urine.

Reaction

Urine is almost always acid when voided, and the more concentrated the urine the higher is the degree of acidity. The acidity is due to the presence of acid sodium phosphate, and, according to some, to free lactic and hippuric acids. Sooner or later, however, the urine becomes alkaline from the decomposition of urea and formation of carbonate and sulphide of ammonium, and deposition of urates. Urine voided before breakfast and during fasting or perspiration is more acid than at other times; during digestion and after meals the acidity is decreased.

Urine may be alkaline when secreted by the kidneys from the administration of fixed alkalies; but urine voided alkaline, unless from this cause, must be due to decomposition of urea and formation of ammonia.

Urine is voided highly acid in acute febrile diseases, especially in acute rheumatism, and it may be voided alkaline after a full meal or after a hot or cold bath.

Alkalinity due to decomposition of urea indicates debility, flatulent dyspepsia, retention of urine, cystitis, etc.

Testing for Acidity or Alkalinity.—To distinguish between alkalinity due to ammonia and to potash or soda, dip a piece of turmeric paper in the urine, withdraw and gently heat it. If ammonia be the cause, the brown colour will disappear; if potash or soda, it will be permanent.

If urine changes a strip of blue litmus paper instantly red, it is highly acid; if slowly to a pink, feebly acid; if not at all, it is neutral or alkaline. If red litmus paper is rendered blue, the urine is alkaline. A quick and easy way to see if urine is neutral is to hang pieces of red and blue litmus paper over the edge of a test-tube into the sample, and if neither is altered in colour the urine is neutral.

Specific Gravity

The specific gravity of urine in health averages at 60° F. 1·020, but varies from 1·015 to 1·025. Specific gravity beads are sometimes used, but the simplest and most usual method is to employ the urinometer. This instrument is used in the following manner: Pour a sufficient quantity of the urine into a cylindrical urine glass, and remove the froth by blotting-paper; then float the urinometer in the urine, taking care that the instrument neither touches the sides nor the bottom of the glass. To correctly read, bring the eye to the level of the liquid, and take the reading from the stem at that part which is on a level

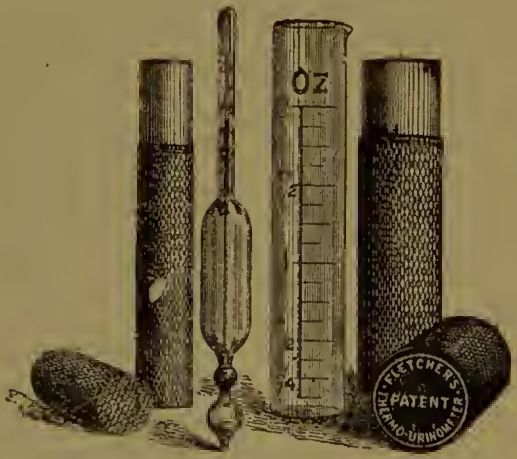

Fig. 1. Fletcher's Thermo-urinometer.

with the surface of the urine. The illustration given on the previous page represents Fletcher's thermo-urinometer, which shows the temperature as well as the density of the liquid.

The variation of the specific gravity of urine depends upon the amount of solids present in it. The subject of total solids is therefore closely connected with specific gravity.

The Solids in Urine

To ascertain the amount of solids in urine, multiply the last two figures of the specific gravity of a twenty-four hours' collection by 2·33. The result gives the amount of solids in 1,000 parts of urine. If, for example, the specific gravity be 1·020, then 20 multiplied by 2·33 shows that a proportion of solid matter, equalling 46·6 parts, has been excreted in every 1,000 parts of urine. So that if the collection during the twenty-four hours amounted to 50 ounces (21,875 grains), the amount of solids can be estimated at somewhat over 2 ounces. Thus :

1,000 : 21,875 :: 46·6 : 2 ounces (roughly).

Another simple method is to multiply the last two figures of the specific gravity by the number of ounces of a twenty-four hours' collection. Thus :

Specific gravity, ·1·019 ; urine collected, 50 ounces.

Then 19 multiplied by 50 would equal 950, which about represents the number of grains of solid matter present in the 50 ounces collected. The quantity of total solids varies in different persons, and in the same persons at different times. According to Beale it may vary from 800 to 1,500 grains, without exceeding the health limit.

Another way of estimating the amount of solids is to evaporate a weighed portion of urine, taken from a twenty-four hours' collection, to dryness, and then weigh the dried product, when a calculation will give the answer.

The amount of organic and inorganic solid matter passed in the urine during twenty-four hours varies very much, depending upon the kind and quantity of food taken, exercise, temperature, abnormal conditions of the body, etc.

According to Oliver the urine of a male adult under normal circumstances should yield from 7 to 8 grains of solid matter per diem for every pound of body weight; women, somewhat less; and children, nearly twice this proportion. After 50 years of age there is a gradual decrease of from 10 per cent. to 30 per cent. in old age. Full feeding and an active life increase the quantity of solids, while reverse conditions decrease the amount.

The urinary solids are deficient when the quantity of urine is normal or subnormal. General causes of deficiency:

1. Habits of life—as light or insufficient diet, sedentary life.
2. Deficiency of muscular tissue, which yields more solids than any other tissue.
3. Defective metabolism of the tissues (which is the cause of the renal inadequacy noted by Sir Andrew Clark).
4. Debility—as in anæmia, hydræmia, cancer, etc.
5. Imperfect action or organic disease of the liver.
6. Acute nephritis.
7. Intercurrent inflamed conditions of chronic renal disease.
8. Hyperæmia of the kidneys, cardiac disease from emphysema dilating the right side of the heart.
9. Early stages of kidney disease.
10. Insidious chronic stage of gouty and tubercular kidney.

The proportion of urinary solids is deficient (quantity of urine increased) in:

1. Diabetes insipidus.
2. Cirrhotic kidneys.
3. Amyloid kidneys.
4. Hydræmia.

The urinary solids are increased when the quantity of urine is normal or subnormal in the following cases:

1. Fevers	Here the percentage of total solids is raised, but the total daily amount is not.
2. Dyspepsia (certain forms) . 3. Lithæmia	In these two examples there is usually a relative increase of urine.

The urinary solids are increased in the following cases :

1. Diabetes mellitus.	
2. Phosphaturia 3. Azoturia (excessive secretion of urea)	These are occasional forms of polyuria depending upon increased metabolism of the tissues.

Qualitative Examination

Set a portion of the urine to be examined in a long urine glass, and after a time decant off the clear fluid, which examine, and subsequently examine the deposit (if any) for the normal and abnormal constituents mentioned in Table II., by the chemical, microscopical, and other tests to be described.

TABLE II.

Abnormal Constituents

Urine free from any well-defined deposit	Albumen Albumoses Sugar Bile acids Bile pigments Hæmoglobin, etc. Urine pigments
Organised deposits	Blood Pus Mucus Casts Micro-organisms.
Unorganised deposits	Urates Phosphates Oxalates Uric acid Cystine Leucine and Tyrosine
Normal constituents present in solution .	Urea Hippuric acid Chlorides Sulphates

Albumen

The first and most important step in the examination of urine for any substance is to precipitate and filter out any albumen, the filtrate and deposit, if any, to be used for the examination of all other substances. Some authorities consider that normal urine should never contain any albumen, while others state that healthy people have occasionally traces of albumen in their urine, due to transitory hygienic, dietetic, or climatic influences, such as biliary disturbances, indigestion, bathing, chills, and sudden changes of the weather.

The morbid condition of the urine known as albuminuria usually arises from mechanical obstruction to the return of the blood to the heart from the kidneys, the kidneys becoming congested and albumen appearing in the urine as long as this congestion continues.[*] If not arrested, this condition usually results in permanent structural injury to the kidneys. In some chronic diseases where there is no mechanical impediment to the return of blood from the kidneys, the presence of albumen in clear urine indicates structural kidney disease. In some acute feverish attacks albumen may appear, but disappears with the illness. Kidney casts should always be looked for in albuminous urine, as their presence indicates that some, if not all, of the albumen is derived from the kidney.

Pus is a common cause of the presence of albumen in urine, and is often present through contact with leucorrhœal or gonorrhœal discharges.

Albumen must necessarily be present if blood is a constituent, being derived from the corpuscles and plasma.

When albumen is present in large quantity, its detection is easy; but when in minute quantities, as in the earlier stages of albuminuria, it is difficult to detect, and may even be overlooked. Decision as to the best time to obtain a sample of urine for the examination of albumen is important, for as the formation of albumen in the urine is usually less during the night, or rest, it follows that morning urine or that collected after exercise will contain more.

Before testing for albumen it is well to see that the urine has an acid reaction, for if neutral, or nearly so, phosphates may be precipitated and interfere with the test.

If any mineral acid be added to albuminous urine before boiling, soluble acid albumen will be formed, and for this reason no albumen will be precipitated. To obtain the acid reaction before testing, it is better to use acetic acid (one or two drops are generally sufficient), as it keeps phosphates in solution and is not so likely to form soluble acid albumen. Urine that has been treated with nitric acid will after a time show crystals of nitrate of urea, but these should be easily distinguishable from an albuminous cloud or coagulum.

Boiling Test.—Filter some urine into a test-tube, and see that there is an acid reaction, by adding one drop of acetic acid. Heat the upper portion of the liquid over a spirit lamp to 212° F. If albumen be present, coagulation ranging from a slight cloud to a dense coagulum will appear in the upper part of the tube, and the unheated lower portion may be contrasted with it. The coagulum is insoluble in acetic or nitric acid. This is the evidence that the coagulum is not due to phosphates. This test precipitates mucin.

Heller's nitric acid test.—To 1 fluid drachm of pure nitric acid in a test-tube add gently 1 fluid drachm of urine; if a more or less dense cloud appears at the junction of the liquids, which is insoluble on heating, albumen is indicated.

Ferrocyanide test.—Acidulate 1 fluid drachm of urine with acetic acid, add solution of potassium ferrocyanide drop by drop till an excess has been reached. If during this addition a precipitate forms, albumen is indicated.

Muter's picric acid test.—Dissolve 7·5 grammes of picric acid in 500 c.c. of water (or 7½ grains to 9 fl. dr.), let it stand some days, and decant off the clear liquid. Now mix equal volumes of filtered urine and reagent in a test-tube, when a white albuminous cloud or precipitate insoluble on heating will be permanent, while that due to peptones, piperazine, urates or alkaloids accidentally present will be dissolved. Picric acid does not precipitate mucus.

Sulphate of copper and caustic potash test.—To some urine in a test-tube add 5 drops of a saturated solution of sulphate of copper and then excess of caustic potash, when a purple colour indicates albumen.

Albumoses

These are bodies closely allied to albumen, and in urine may appear as peptones, globulin, etc.

Peptones are formed during the digestion of flesh in the stomach ; they remain in solution, and, unlike other varieties of albumen, they are not precipitated by heat, acids, or alkalies. Peptones are chemically related to albumen, in much the same way as starch is to sugar. In addition to the tests by heat, nitric acid, and picric acid, their presence can be detected in using Trommer's and Fehling's sugar tests. With Trommer's test, if a rose-red or pink tint appears on adding the liquor potassæ to the urine and copper solution before heating, peptones are indicated. In the same way this colour is found at the junction of the urine with Fehling's test solution.

Peptonuria may be present in local inflammatory affections acute and subacute nephritis, pneumonia, pleurisy, diphtheria, typhus, typhoid, small-pox, etc.

Globulin is a substance which may be obtained from blood serum, and is chemically allied to albumen, but, unlike albumen, it is insoluble in water, and soluble in urine on account of the saline matter contained in it. It is, however, precipitated by heat and nitric acid. Globulin can be obtained from urine by diluting it with water, when it falls out of solution, rendering the urine milky ; it can then be filtered out. Globulin may be present in acute nephritis, acute Bright's disease, waxy kidney, and catarrh of the bladder.

Glycosuria

Grape sugar in small quantities is sometimes found in urine as a constituent in health, but it occurs in abundance in the disease known as diabetes mellitus, when a very large quantity of urine is passed, the number of ounces daily being often increased to as many pints. It is said that healthy urine may contain as much as ·01 per cent. of sugar, or about 15 grains daily, and no doubt many people pass sugar who have no diabetes—the glycosuria of the middle-aged, free-living, gouty, and plethoric being examples of this. Whilst the excretion of urea and albumen is said to be greater during the day, the secretion of sugar is found to be greater during the night; hence a morning collection of urine should contain more sugar. If sugar is present in considerable quantities at periodical examinations, diabetes mellitus may be diagnosed; but if the amount is small and variable, there may not be any disease present.

In glycosuria the kidneys become enlarged and congested, and the functions of the liver are impaired. In health the portal circulation carries off from the stomach starchy matters, which are taken to the liver and converted into glucose. This glucose, after elaboration in the hepatic cells, is then conveyed (as hepatin) through the hepatic vessels, the inferior vena cava, and the right side of the heart to the lungs, where, meeting with the oxygen of the air, it is consumed, carbon dioxide and water being given off. In diabetes mellitus this does not occur, for after exposure to the air in the lungs it passes on to the left side of the heart, containing some or all of the sugar which is subsequently found in the urine.

Some authorities on diabetes mellitus consider the disorder due to brain disease. These, therefore, do not accept the theory we have stated.

Sugar is present in the urine during the convalescence from some acute disorders, and especially in cholera, carbuncle, and certain injuries to the nervous system.

Certain drugs—such as chloral, phenacetin, morphine, and chloroform—like sugar, reduce the cupric salt in Fehling's solution to cuprous oxide; uric acid in excess, mucus, and some extractives also do the same.

Fehling's test solution, when kept some time, often deposits cuprous oxide on heating (without urine being present); therefore it should be freshly made, or boiled to see if the reagent is right, before using.

The specific gravity of saccharine urine varies from 1·020 to 1·060; but traces may be found in urine having a less specific gravity than 1·020.

A form of diabetes called diabetes insipidus, properly known as polyuria, is not accompanied by sugar. The urine in these cases has a specific gravity ranging from 1·001 to 1·010. It is a curious and rare disorder, the quantity of urine passed being often as great as in true diabetes, pailfuls being passed daily, and the patient has a constant thirst and desire to pass water. This disorder is supposed to be to a great extent hereditary; some sufferers live to a good old age, whilst others fail in health, death generally occurring from some other complaint.

Tests for Grape Sugar

These tests are to be applied after removing the albumen (if any).

Moore's test.—Mix equal quantities of urine and liquor potassæ in a test-tube, heat to 212° F., and boil for a minute or two. If sugar be present, the urine will turn dark red, brown, or even black; whilst non-saccharine urine would only turn a brownish-red.

Trommer's test.—To 1 fluid drachm of urine in a test-tube add 10 drops of a solution of sulphate of copper (10 grains to the ounce), then add liquor potassæ drop by drop until the precipitate first formed is re-dissolved, slowly heat the solution to 212° F., when, if sugar be present, an orange-red precipitate, after a time becoming reddish-brown, is thrown down.

If a similar solution, not heated but set aside in the cold, gives at the end of twenty-four hours a reddish precipitate, sugar is undoubtedly present.

Fehling's test.—This test is generally kept in two solutions—solution No. 1 containing sulphate of copper and water; and solution No. 2 caustic soda, tartarated soda, and water. When wanted for use they are mixed in equal volumes, each 10 c.c. o the mixed test requiring ·05 gramme of glucose to discharge the blue colour and give the cuprous oxide precipitate.

The principle of the estimation of sugar by Fehling's test is that if a mixture of urine, 10 c.c. diluted to 100 c.c. with recently boiled water, is allowed to run gradually from a burette into 10 c.c. of Fehling's solution—the mixture meanwhile being heated until the blue colour is discharged—the number of c.c. of urine used will contain ·05 sugar, and the sum will then work out thus : $\frac{100 \times \cdot 05}{\text{c.c.}} =$ in gramme the amount of sugar in the 10 c.c. of urine taken.

Before testing or estimating with Fehling's solution it is well to precipitate and filter out phosphates, as sometimes with Trommer's and Fehling's tests a dirty-looking pinkish deposit is met with, due to phosphates and black oxide of copper mixed, the phosphates being tinted pinkish by urine pigments.

Roberts's fermentation test.—After collecting and measuring the urine passed in twenty-four hours, take two 8-ounce bottles; into one put 4 ounces of urine and compressed yeast about the size of a chesnut, and into the other 4 ounces of urine only. Cork both bottles well, and allow them to stand twenty-four hours in a warm place; say, on the hob or mantelshelf; then pour the contents of each bottle into separate urine glasses, and take the specific gravity of each, when the difference of each degree lost in the urine containing the yeast indicates the presence of 1 grain of sugar in every ounce. For example, if the specific gravity of the unfermented urine is 1·040, and that of the fermented 1·010, the urine contains 30 grains of sugar in every fluid ounce; and if the number of ounces of urine passed in the twenty-four hours be multiplied by the number of grains in an ounce, the result is the amount per diem.

There are other more elegant and perhaps more accurate methods of performing this test; but as far as the qualitative analysis is concerned, if fermentation sets in and the specific gravity is decreased to any extent, this test may be considered simple and trustworthy.

Picric acid test.—Take a test-tube half full of urine and add 1 fluid drachm of saturated solution of picric acid and 1 fluid drachm of liquor potassæ. Boil. If sugar be present, the yellow picric acid is converted into the red picramate of potash, the depth of colour indicating the strength of sugar.

It has been thought that the coma which so frequently terminates life in diabetes is due to the presence of acetone in the blood. When this is the case, its presence is shown by the urine giving a deep red coloration with perchloride of iron.

Dr. Ralfe's test for acetone.—Take 1 fluid drachm of liquor potassæ in which 20 grains of potassium iodide have been dissolved, and boil; while still hot pour 1 fluid drachm of urine on the surface of the test liquid, when at the line of contact a ring of phosphates will appear. If acetone be present, this ring becomes yellow in a few minutes, and numerous points of iodoform appear, which finally sink to the bottom of the test-tube.

Bile

Urine is of a greenish-brown colour when it contains bile (the depth of colour depending upon the amount of bile present), and it colours white filtering paper yellow. In examining urine for bile, there are two sets of bodies which, if found, will prove its existence, viz. bile acids and bile pigments.

Pettenkofer's test for bile acids.—Mix urine with half its bulk of strong sulphuric acid in a test-tube (rise of temperature being prevented by immersing the tube in water). A small quantity of white sugar is then added and well mixed with the acid and urine; more sulphuric acid is now added, and as the temperature rises a reddish or violet coloration is produced, the liberated cholalic acid due to the decomposition of the taurocholate and glycocholate of soda furnishing the colour.

The presence of albumen or volatile oils interferes with this test.

Gmelin's test for bile pigments.—Into a test-tube containing the depth of half an inch of nitric acid (commercial acid containing free nitrous acid is best) and held at an angle, pour gently a little urine down the side of the tube, so that the two fluids may touch, but not mix. At the line of contact a zone of red appears in every sample of urine, but, if pigment be present, a zone above becomes first green, then violet, blue, and red, representing the various stages of oxidation of the pigments; but the most characteristic colour, and indeed the only reliable one, is the green. Traces of bile pigments and acids are present in the urine at all times, and are increased in quantity during warm weather, but if in excess they indicate liver disease or phosphorus poisoning.

Methyl-violet test for bile pigments.—Pour a solution of methyl violet down the side of a test-tube to form a layer at the bottom of some urine previously poured into the tube, when a carmine-red ring will appear at the junction of the two liquids.

Marechalt's iodine test for bile pigments.—A trace of tincture of iodine colours the urine green. The same test is recommended by Rosin.

Strasburger's test for bile acids.—Mix some urine with ordinary cane sugar, and saturate a piece of filtering paper with it. Touch the paper with a glass rod dipped in strong sulphuric acid, when a purple spot denotes bile acids.

Precautions in Testing for Bile Pigments

Indican.—This is the only substance likely to be mistaken for bile pigments, as Gmelin's test may give a red, violet, or even a green colour with it. Indican exists in small quantities in normal urine, but in excess in wasting diseases, such as phthisis, cancer of the liver or stomach, Addison's disease, cholera, and typhoid fever. Decomposition sometimes takes place in the bladder, when the urine is voided blue. After passing urine, where this decomposition has not taken place it can be accomplished by mineral acids as follows: Mix equal parts of strong sulphuric acid and urine; if the urine becomes black and opaque, depositing a purple precipitate on diluting with water, indican may be suspected and proved by the following corroborative test : Mix equal parts of hydrochloric acid and urine, add two or three drops of a saturated solution of chlorinated lime, when, if indigo be formed, colouring the mixture blue, indican is present. This test distinguishes between bile pigments and indican.

Santonin colours urine yellowish or greenish, and may be mistaken for bile pigments. The following test will distinguish between them : If caustic potash be added to urine containing santonin, a reddish-purple colour appears, which becomes bleached by an acid.

Iodides or Bromides in the urine may on the addition of nitric acid and heat liberate free iodine or bromine.

Aniline.—Preparations of aniline, as fuchsin, may colour the urine.

Hæmoglobin

The colouring matter of the red blood corpuscles, having escaped from them, may appear in the urine, thus altering the colour and forming the condition known as hæmoglobinuria. If no corpuscles are present in the urine, which is most frequently the case in hæmoglobinuria, the task is more difficult, and hæmoglobin or hæmatin must be searched for, the presence of one proving the presence of the other. Hæmoglobin contains ·04 per cent. of iron, and hæmatin about 9 per cent.

Hæmatin test.—Precipitate the phosphates from the urine by caustic potash and gentle heat, dry the phosphates on an object glass, add a crystal of common salt, and place on it a cover glass. Warm the preparation, then cool and examine by the microscope for hæmatin, in small reddish-brown acicular crystals insoluble in water, but soluble in caustic potash.

Guaiacum test.—Add to the urine a few drops of freshly made tincture of guaiacum and some ethereal solution of peroxide of hydrogen, when, if blood be present, a fine sapphire-blue colour is developed. Hæmaglobinuria occurs in most cases of jaundice, in poisoning by sulphuric acid, phosphorus, and arseniuretted hydrogen, and in scurvy.

The Pigments of the Urine

Urochrome.—This is always present, and produces the normal yellow colour of urine.

Uro-erythrin (purpurin).—This is always present, and gives the characteristic pink colour to urates and probably to uric acid.

Melanin.—The urine of patients suffering from melanotic cancer is often of a dark brown colour, from the presence of this pigment.

Blood

Is frequently met with as a deposit in urine, and may be derived from any part of the urinary-renal tract. If derived from the kidneys, it is generally diffused through the urine ; if from the prostate or bladder, it is in excess at the end of micturition ; and if from the urethra, the greatest amount is in the beginning of micturition. The appearance of the blood also indicates at what part the hæmorrhage exists—the colour ranging from a characteristic smoky appearance, if from the kidneys, to blood-red if from the urethra.

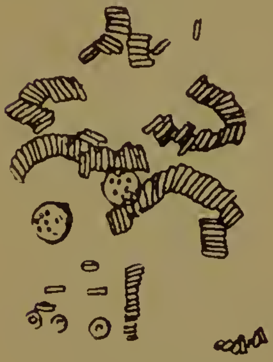

Fig. 2.—Blood Corpuscles.

In examining blood corpuscles under the microscope the peculiar colour and circular disc-like appearance of the red corpuscles distinguish them from any other deposit. When these blood corpuscles, containing their colouring matter hæmoglobin, are present in the urine, the condition is known as hæmaturia, and indicates :

Acute Bright's disease.
Congestion of the kidneys.
Cancer of the kidneys.

External injury to the kidneys.
Calculus in pelvis or ureter.
Calculus of the bladder.
Cancerous growth of the bladder.
Congestion or ulceration of the mucous membrane of the bladder.
Mechanical injuries to the urethra.
Gonorrhœa.
Menstruation, etc.

In examining urine containing blood, albumen is always present in proportion to the quantity of blood, and may be so abundant that the urine will solidify on heating. Such urine soon becomes alkaline; therefore the acid reaction should be restored before testing for albumen. If the blood is considered to be from the kidneys, search should be made for renal casts; if from the pelvis or ureter, there may also be pus, and possibly gravel.

Examination Tests.—1. Coagulability by heat and nitric acid.

2. Urine albuminous in proportion to the quantity of blood present.

3. Perhaps presence of pus and gravel, if from the pelvis of kidney or ureter.

4. Solution of ammonia gives to blood a higher crimson tint.

Van Deen's Test.—Add freshly prepared tincture of guaiacum and an ethereal solution of peroxide of hydrogen to a portion of the urine, when, if a sapphire-blue colour is produced, the presence of blood may be suspected. Saliva, nasal mucus, and potassium iodide give the same reaction; hence the microscope and spectroscope tests are the best.

Microscope.—The blood corpuscles do not form rouleaux, and may be either biconcave or biconvex, regular or irregular, yellowish or colourless, or absent as in hæmoglobinuria.

Spectroscope.—Solutions of blood give rise to two dark lines, called absorption bands, situated in the green and yellow of the spectrum between the D and E lines of Fraunhofer.

Pus

Is often present in the urine, forming a thickish creamy-looking deposit, the supernatant fluid being somewhat turbid from the presence of some diffused pus corpuscles. The presence of pus may be due to any of the following causes :

Leucorrhœa.
Gonorrhœa.
Pyelitis.
Cystitis.
The bursting of an abscess in any part of the urinary tract.
Gout (frequent).

In the examination for pus the following are the chief points :

Urine containing pus soon becomes alkaline from the decomposition of urea, the pus changing into a gelatinous stringy mass adhering to the sides of the vessel.

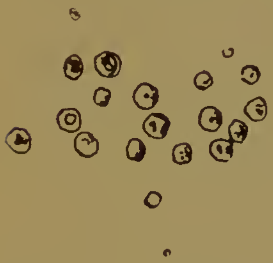

FIG. 3.—PUS CORPUSCLES.

The action of liquor potassæ upon pus corpuscles produces the same effect as if from the decomposition of urea.

Albumen derived from the corpuscles is always present with pus, and kidney casts should be looked for.

Under the microscope the corpuscles are rounded and colourless, having granular contents with from one to four nuclei, these nuclei being seen more clearly after treatment with acetic acid. In size the pus corpuscle is somewhat larger than the blood corpuscle.

Mucus

Is a deposit consisting chiefly of epithelial débris. In healthy urine there is always a cloud present, but in catarrhal and inflammatory conditions of the genito-urinary passages mucus exists in excess. It is distinguished from pus as follows :

Dilute acetic acid precipitates mucin and not pyin.

Mucus, unlike pus, does not soon lose its acid reaction.

Mucus does not mix freely with urine ; pus does.

Mucus does not necessarily accompany albuminous urine ; pus does.

Mucus does not show definite corpuscles under the microscope ; pus does.

Mucus is not converted into a stringy, ropy tenacious mass by liquor potassæ ; pus is, and mucus is rendered more fluid.

The part affected is indicated by the epithelial cells seen under the microscope :

Spheroidal single and multiple nucleated epithelium comes from the kidneys. Irregularly columnar nucleated epithelium indicates that the pelvis, ureter, or prostate is involved.

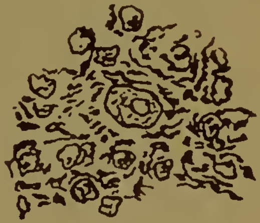
FIG. 4.—MUCUS DEPOSIT.

Irregularly circular squamous nucleated epithelium is derived from the bladder, urethra, or vagina.

Casts

A cast is a mould of any part of a uriniferous tube, formed there and discharged by the urine. These can scarcely be overlooked if the urine is allowed to settle for a few hours in a tall cylindrical glass, the whole of the supernatant fluid decanted off, and the last drops put under the microscope for examination. Or, to be more exacting, a quantity of urine should be used, and several hours allowed for it to settle ; then decant off all but 4 ounces, allow this to settle, and again decant, using the last few drops for the microscope. It sometimes happens that urine may contain casts, but, the specimen sent being an upper stratum of what was in the utensil, the casts have been left behind. If therefore casts are suspected, the entire contents of the vessel should be obtained for examination. These casts are probably formed in the straight uriniferous tubes by the escape of blood or plasma into them, and by the subsequent coagulation of the fibrin the whole becomes moulded to the shape of the tube. According to others they are epithelial and granular casts produced by the desquamation and degeneration of renal epithelium. Sometimes foreign bodies are thrown into urine by accident or purposely, as cotton fibre, sputa, etc., but practice with the microscope will enable the analyst to detect these readily.

Kidney casts vary from $\frac{1}{3000}$ to $\frac{1}{300}$ of an inch in diameter, and rarely exceed $\frac{1}{50}$ of an inch in length. They are more or less cylindrical in form, varying little in diameter throughout, and are not twisted as many foreign matters, such as hair or cotton fibre might be. The usual kidney casts are classified as follows :

Blood cast.—A cast composed of fibrin formed by the coagulation of blood plasma, and exuded from the surrounding capillaries into the uriniferous tubules, indicating an escape of blood from the surrounding vessels, as in acute Bright's disease, strangury, and hæmaturia.

Pus cast (purulent cast).—Indicates coagulation of escaped pus entangled in casts of fibrin, as in renal abscess or suppurative nephritis.

Hyaline casts (transparent or waxy).—Varieties :

Epithelial.—When the epithelial calls are loosened and surround a fibrin cast.

Granular.—When the loosened cells have undergone disintegration, having a dark and granular appearance, occurring after frequent attacks of gout and being about $\frac{1}{700}$ of an inch in diameter.

Fatty (oily).—When the disintegrated cells have undergone fatty degeneration, in which case entangled oil globules and epithelial cells gorged with oil indicate fatty degeneration of the kidneys.

Crystal-containing.—Hyaline cells containing crystals of oxalates and phosphates.

Micro-organisms

The term micro-organism is given to certain living organised beings found almost everywhere ; in the air, in water, in the soil, in the alimentary canal or in any liquid (not poisonous to them) containing organic matter upon which they can live, such as milk, blood, pus, mucus, urine, etc. They occupy such a low position in the organic world that there still exists a difference of opinion as to their proper place in it. It is, however, agreed that a majority belong to the lower fungi or possibly algæ. They vary in size, the smallest being about ·0011 mm. in diameter and about twice that in length, so that with a microscope having a power of 3,000 they look little larger than the dots or commas of ordinary type. The micro-organisms commonly met with in urine are :

1. **Micrococcus ureæ**, by which urea is decomposed into ammonium carbonate.

2. **Micrococcus albicans amplus** (Bumm), which, if found, indicates the presence of leucorrhœal discharge.

3. **Gonococcus**, which, if found, indicates the presence of gonorrhœal discharge.

4. **Streptococcus pyogenes**, which, if found, indicates the presence of pus.

N.B.—All the coccus forms of micro-organisms are composed of round or ovoid cells, isolated or in chains, or in gelatinous families adherent by a mucilaginous secretion developed around each cell.

5. **Sarcinæ.**—These are square little bodies formed in the urine before it is voided, and are similar to the sarcinæ found in the vomited matters of persons suffering from stenosis of the pylorus. They belong to the coccus form of micro-organisms, being composed of round or ovoid cells, which develop by gelatinous adherence into square-like bodies arranged in fours or multiples thereof.

6. **Vibrio.**—When urine (especially albuminous) has been allowed to stand, little elongated bodies invariably appear consisting of filaments with numerous transverse lines. These micro-organisms exhibit active movements, the longer ones twisting about in various ways. They are of the bacterium form, being composed of rod-like cylindrical cells, isolated, interlaced, or in bundles, and may be wavy or spiral.

7. **Torula cerevisiæ.**—This is the yeast which induces alcoholic fermentation, and, if present, indicates glycosuria. The organism is composed of oval cells joined together in a row, each cell being about the size of a blood corpuscle.

8. **Penicillium glaucum** (common green mould).—This is occasionally observed in urine which has been kept a long time, but is without significance to the physician.

Various other organised constituents may be found in the urine, such as bladder or urethral epithelium casts of the seminal tubes (which are much longer than those of kidney tubes), casts of the uterus or vagina, vaginal epithelium, etc. The detection and identification of these organised constituents require much practice, and often puzzle the most experienced. If an analyst has, however, any doubt as to the nature of such a constituent, rather than wrongly name it, it is better to sketch and describe it in a footnote to the report.

Spermatozoa

Are present in the urine first passed after an emission of semen, and when found in the urine of women are proof of sexual intercourse, the seminal secretion appearing as a white glairy deposit, showing under the microscope spermatozoa having the characteristic oval head and long fine whip-like tail—the tail, however, losing the movement it has in urine.

These are liable to escape notice on account of their small size and extreme transparency. Suspected urine should be allowed to stand some hours in a conical glass, and the drop at the bottom examined with a microscope of high power, when the tadpole-like appearance of the spermatozoa, if present, renders them easily recognisable. Spermatozoa may be seen with a microscope having a power of 250 diameters.

Urinary Deposits

In examining the unorganised deposits, the first four in the table—viz. urates, phosphates, oxalates, and uric acid—can almost be distinguished by the naked eye from the other three. By their appearance, and by chemical and microscopical examination, they may be readily distinguished as follows :

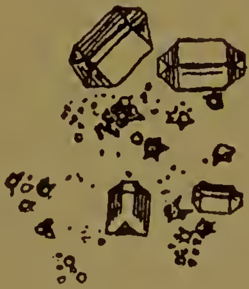

Fig. 5.—A Urinary Deposit.

Urates

Warm the urine containing the sediment. If the latter dissolves and the urine becomes quite clear, the precipitate again falling when the urine cools, the sediment consists entirely of urates in combination with sodium, ammonium, or calcium. The microscopical appearance shows what the urates are.

Urate of soda is the most frequent of the three salts, and is usually an amorphous deposit, but occasionally occurs as little dark bodies with short spikes.

Urate of ammonia is rarer, and occurs in a globular form with short projecting spikes like the soda salt.

Urate of lime is very rare, and forms only an amorphous deposit.

If in doubt about the nature of these salts, add a drop of hydrochloric acid to the specimen, when crystals of uric acid will be formed. If still in doubt, apply the murexide test with the crystals as follows : Place a portion of the crystals in a small Berlin porcelain dish, remove moisture by bibulous paper, add a drop or two of strong nitric acid, and evaporate to dryness at a gentle heat ; when cold, add a drop of solution of ammonia, which produces a purple colour, deepening to violet on adding a drop of liquor potassæ.

Urates differ in colour considerably, according to the amount of colouring matter in the urine. They are usually seen as a brick-dust deposit, the colouring matter around them receiving the name of purpurin; but in young children there is sometimes seen milky urine, due to a deposit of white urates. It is generally considered that urine contains one part of urates in a thousand, and when in excess of this the salts are deposited as a sediment, but only after passing and when the urine has cooled. This deposit is the most common and least important of all urinary sediments.

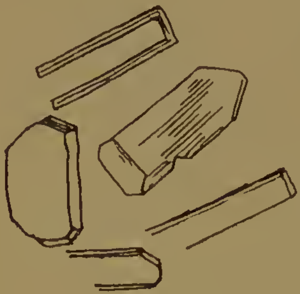

FIG. 6.—UREA CRYSTALS.

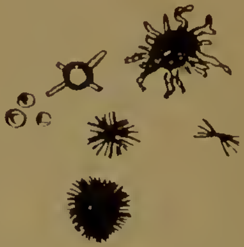

FIG. 7.—ACID SODIUM URATE.

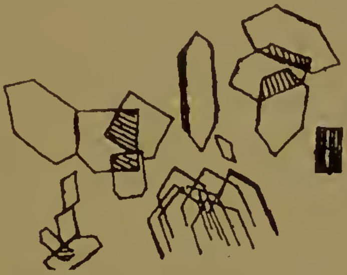

FIG. 8.—UREA NITRATE.

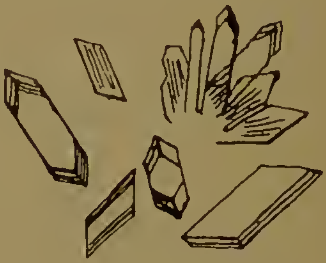

FIG. 9.—UREA OXALATE.

Phosphates

If the deposit does not dissolve by heat, let it settle, wash once by decantation with cold water, and warm with acetic acid, which will dissolve all earthy phosphates; now throw down the phosphates from the solution by solution of ammonia, filter out, wash the filter with boiling water, and dissolve the deposit in acetic acid, and test (if necessary) for calcium and magnesium. After filtering out earthy phosphates from the urine, the alkaline phosphates can be precipitated by magnesia mixture as $MgNH_4PO_4$.

The microscopic appearances of phosphates occurring as deposits in urine are as follows :

Ammonio-magnesium phosphate appears under the microscope as right rhombic prisms soluble in acetic acid (distinction from oxalates).

Phosphate of lime occurs as an amorphous precipitate, but occasionally in stellate grains known as stellar phosphate.

Phosphoric acid is contained in urine in combination with potassium, sodium, calcium, and magnesium. Its combinations with the former two bases constitute the alkaline phosphates, and with the latter two the earthy phosphates. Earthy phosphates are deposited in the urine when neutral or only faintly acid, and may be deposited before or after the urine has left the bladder, the deposition depending upon whether decomposition of urea sets in before or after voiding the urine.

When urea has undergone ammoniacal decomposition, either in the body or after the urine is passed, an additional earthy phosphate occurs, viz. the ammonio-magnesium phosphate, which is not a normal salt of urine, but one depending upon the alkalinity due to this decomposition of urea. When these earthy phosphates are deposited in the bladder as neutral phosphates, they are passed at the end of micturition, looking like a short stream of milk. Phosphates of lime and magnesia are insoluble in water, but are kept in solution in the body as acid phosphates ; so that, if healthy urine is neutralised by the administration of fixed alkalies, phosphates may be formed in the bladder and passed in the urine.

The presence of phosphates as a deposit in urine, known as phosphaturia, indicates fevers, disease of the nerve centres and of bone, dyspepsia, and Bright's disease. They always indicate depression and debility, and, if in excess, waste of tissue.

Calcium Oxalate

If the deposit is insoluble in acetic acid, warm another portion of the urine with hydrochloric acid. If soluble, the deposit is calcium oxalate, and may be re-precipitated by ammonia. The blowpipe will convert it into carbonate, which may then be dissolved in weak acetic acid and tested for calcium with ammonium oxalate.

The microscope shows oxalate of calcium as square white octahedra, or as dumb-bells without fringed extremities.

The presence of this salt in the urine constitutes a condition known as oxaluria, but does not indicate anything serious. Oxaluria is usually associated with gastric disorders. Crystals of uric acid are often found in the urine in oxaluria. Oxalate deposits never occur in any great quantity, for in oxaluria the urine excreted in twenty-four hours seldom contains more than two-thirds of a grain of oxalate of calcium.

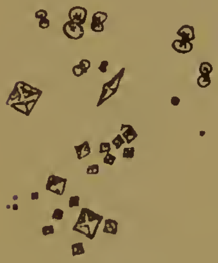

FIG. 10.—CALCIUM OXALATE.

Uric Acid

If the deposit is insoluble in hydrochloric acid, test the urine for uric acid by the murexide test described under urates.

The microscope shows crystals of uric acid as follows:

1. Dumb-bell crystals with fringed extremities.
2. Rhomboidal, or long oval plates with acute angles; such crystals being often interwoven, looking like rosettes.
3. Rectangular, barrel-shaped, or in hexagonal plates with two parallel sides longer than the other four.

Uric acid generally exists in urine in combination with sodium and potassium, but it may be set free in disease, and is then seen in the urine like grains of cayenne pepper or coarse sand adhering to the sides of the vessel. The colour of the crystals is really white, but they appear to be pink, the colour being derived from the urine pigments.

Such deposits often precede glycosuria, and may alternate with it. Crystals of uric acid are apt to form in the kidneys or bladder; and in those cases, if they are detached and pass off with the urine, they are called gravel; but if they are attached and do not pass off with the urine, a calculus forms. Uric acid may be secreted directly by the kidneys, or may be due to the decomposition of urate of ammonium in the presence of some free acid.

Urine containing free uric acid is generally very acid, of a dark colour, of a high specific gravity, and the quantity voided is under the average. It is important not to mistake the brick-dust deposit of urates for uric acid, as they are never present except in solution) in the kidneys or bladder at the normal temperature of the body. Uric acid does not affect the appearance of the urine; the crystals simply roll to the bottom or adhere to the sides of the vessel, the urine otherwise looking clear and normal.

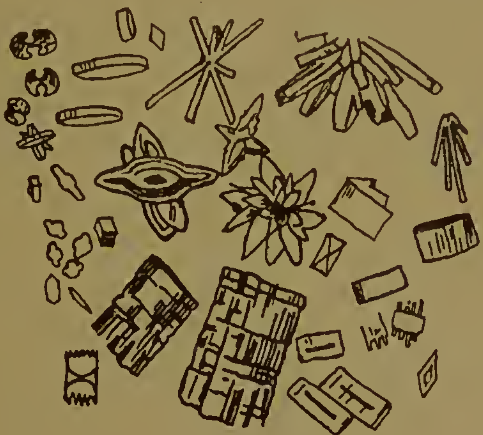

FIG. 11.—URIC ACID.

If, however, there should be reason to suspect a mixed deposit of uric acid and urates, heat will dissolve the urates, which can then be separated from the uric acid, thus permitting the examination of both. The tendency of urine to contain free uric acid occurs most frequently in dyspeptics of gouty habit. Such a condition is known as the lithic or uric acid diathesis.

Uric acid is really a less oxidised stage of urea. It is found in the urine of all carnivorous animals, and in reptiles it entirely replaces urea. To ascertain if urine contains free uric acid, acidulate an ounce with a dram of strong hydrochloric acid in a beaker. Cover the top, and set aside for a day or two, when, if uric acid be present, the crystals will be seen on the sides and bottom of the beaker, or on the surface of the fluid.

The amount of free uric acid in urine is increased when there is an increase of urea, as in acute rheumatism and chronic liver diseases, and is often absent from urine before an attack of gout, and may even disappear for some days during the attack. The solubility of uric acid being only 1 in 15,000 of cold water and 1 in 1,900 of hot water (urates being somewhat more soluble) accounts for the commonly-met-with urate deposits.

Cystin,

also called cystic oxide, is a rare organic deposit composed of carbon, hydrogen, and nitrogen, with a large quantity of sulphur. It is thought by some to be a derivative of albumen. It consists of regular colourless hexagonal plates, superimposed, and is distinguished from the hexagonal plates of uric acid by absence of colour, or a yellowish-green tint only. Cystin is sometimes found mixed with uric acid and urates. Urine containing cystin is feebly acid, of a yellowish-green tint, and of a sweetbriar odour (sometimes a putrid cabbage odour), urea and uric acid being present as a rule in a diminished quantity.

In addition to the microscopical examination just mentioned, the following chemical test should be applied: Boil a portion of the urine containing the deposit with caustic potash and acetate of lead, when a black precipitate is formed, due to the formation of black sulphide of lead by the action of the sulphur of the cystin. This test, known as Liebig's, is, however, not quite reliable, as other things, especially albumen, contain enough sulphur to give the reaction. Cystin when dissolved in urine may be thrown down by acetic acid and the precipitate examined under the microscope.

Cystin is soluble in solution of ammonia, but is precipitated unchanged upon the spontaneous evaporation of the ammonia.

Solution of nitro-prusside of sodium gives a violet colour if added to an alkaline solution of cystin. The tendency to formation of cystin in urine is by some considered to be hereditary, but the substance has been found in the urine of chlorosis and some calculous disorders.

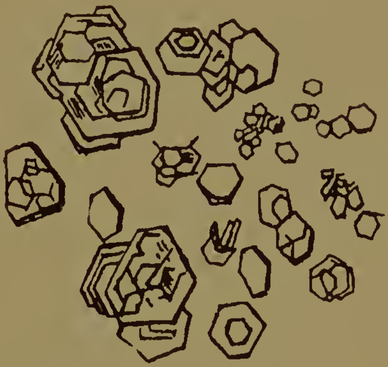

FIG. 12.—CYSTIN.

Leucin and Tyrosin

These are very rare deposits, and are usually found associated in acute yellow atrophy of the liver, and frequently in small-pox, typhoid fever, and acute tuberculosis.

Tyrosin crystallises in bundles of shining delicate needles, the bundles being sometimes arranged in a stellate form.

Leucin appears in gland-like masses, circular oil discs, or dark globular crystals. These bodies do not occur as a deposit. They may be looked for by Hoffman's test. A large quantity of the urine is fully precipitated by a solution of acetate of lead. Through the filtrate pass sulphuretted hydrogen, to remove excess of lead; evaporate the filtrate to a syrupy consistency, and then extract with alcohol. The leucin will be in the alcoholic solution, but the tyrosin will not. Dissolve the part insoluble in the alcohol in water, and boil with a few drops of nitrate of mercury solution, when, if tyrosin be present, the fluid becomes rose-red, and finally a red precipitate is thrown down.

The alcoholic solution containing the leucin is now evaporated to dryness and dissolved in ammonia solution ; acetate of lead solution is then added as long as a precipitate continues to form. The leucin, in combination with lead, is now washed on a filter, and afterwards diffused in water through which a stream of sulphuretted hydrogen is passed. The liquid is again filtered, and the filtrate evaporated and set aside to crystallise. The resultant crystals are tested as follows : They are heated with nitric acid on a platinum crucible, when, if leucin be present, a colourless, almost invisible, residue is left, which, warmed with a few drops of soda solution, becomes yellow, changing to brown.

Another test is that if leucin be heated in a dry test-tube oily drops are formed, giving off the smell of amylamine. Again, leucin placed on platinum foil with a drop of nitric acid gives a colourless residue, which on the addition of caustic potash and further heating yields yellow-brown oil drops.

Crystals in Urine

The recognition of crystals under the microscope having the same appearance as leucin or tyrosin cannot be depended upon, as in health and disease crystals identical in form are sometimes found in urine which are neither of the substances in question. In addition to the normal constituents present in abnormal quantities as deposits, and abnormal constituents already referred to, there are other normal constituents of the urine which may be present in solution in abnormal quantities—that is, either more or less than normal—the quantitative analysis of which will be treated later. These constituents are urea, hippuric acid, chlorides, and sulphates, the most important being :

Urea.—In a healthy man from 300 to 500 grains of urea are passed with the urine in a day. The amount is increased by a meat diet, and diminished by a vegetable one. An increase also occurs during febrile attacks and from tissue metamorphosis ; while in some diseases, such as uræmia and Bright's disease, it is below the average. For recognition and isolation see section under Quantitative Analysis (p. 42).

Hippuric acid is excreted in quantities varying from 7 to 15 grains per diem, chiefly as hippurates of soda and potash. It is increased by eating much fruit, especially plums, blackberries, and cranberries ; or by the administration of benzoic acid during health. In disease benzoic acid when given internally is said to be eliminated as hippuric acid. The formation of hippuric acid from benzoic acid is caused by the action of glycocol in the kidneys. The amount of hippuric acid in urine is increased in diabetes.

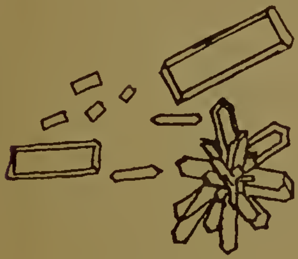

FIG. 13.—HIPPURIC ACID.

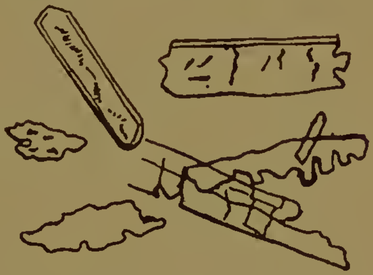

FIG. 14.—BENZOIC ACID.

The tests for hippuric acid are :

1. A neutral solution of ferric chloride gives a brownish precipitate on boiling.
2. Hydrochloric acid added to urine in excess gives on boiling a white precipitate of benzoic acid.
3. Evaporate 15 ounces of urine to 3 ounces, acidify with acetic acid, and stir in sufficient plaster of Paris to make the whole set firmly. When set, powder and extract the hippuric acid with ether, evaporate the ethereal solution, and, when all the ether has passed off, the pure hippuric acid will be left in long needle-shaped crystals. Place a few crystals in an evaporating dish, add a drop of nitric acid, evaporate to dryness, and heat the residue to redness on platinum foil, when a smell like oil of bitter almonds will be given off.

Chlorides are excreted as chloride of sodium in quantities averaging about 250 grains in twenty-four hours. This amount is diminished, and may even entirely disappear, in acute pneumonia, acute rheumatism, and most febrile diseases. In disease the amount of chloride depends generally upon the digestive powers of the patient. Chlorides can be shown by acidulating the urine with nitric acid (to keep the phosphates in solution), and adding a solution of silver nitrate as long as a precipitate falls; the precipitate is silver chloride, which is soluble in solution of ammonia, but is re-precipitated by nitric acid.

Sulphates are increased by an animal diet, but their presence is not considered of much importance. Normal urine contains from 1·5 to 3 parts per 1,000 of SO_3. Sulphates may be precipitated as barium sulphate by first acidulating the urine with hydrochloric acid, and adding solution of barium chloride as long as a precipitate continues to fall.

QUANTITATIVE ANALYSIS

1. Albumen, sugar, urea, uric acid, chlorides, phosphates, sulphates.
2. Examination of sugar by the polariscope.
3. Examination of blood by the spectroscope.
4. The introduction of foreign matters into urine accidentally or by design.

Preliminary Preparations

The urine passed during twenty-four consecutive hours should be obtained from 9 A.M. one day to 9 A.M. the next day, this being the best time to get a good average result. The patient should pass water just before 9 A.M. the first day, but this portion should not form part of the collection. The last portion, however, passed at 9 A.M. the second day should be included. The urine collected must be kept in a clean clear-glass stoppered bottle.

When the analysis is to be made, the urine should be measured in a litre measure graduated with divisions not exceeding 10 c.c. If albumen be present, it must be removed before examining for any other constituent. Thus, heat 200 c.c. to boiling-point in a Berlin porcelain dish, the bottom of which is protected by a piece of wire gauze. If the albumen do not separate in flakes, dilute acetic acid may be carefully added until the acid reaction is well marked. The analyst must be careful not to add too much acetic acid, or part of the albumen may be re-dissolved. The heat must be moderate, and must not exceed boiling-point, for if too high the urea may be decomposed. The urine is then filtered, and put into the measure in which the 200 c.c. was measured. The dish and filter are then washed with sufficient distilled water to make up the original quantity. The fluid is now ready, without further preparation, for the estimation of sugar, urea, uric acid, chlorides, phosphates, sulphates.

The metric weights and measure usually employed in quantitative analysis are:

The litre (1·76 pint), the c.c. or cubic centimetre (the $\frac{1}{1000}$th part of a litre), the gramme (15·43 English grains), and the milligramme (the $\frac{1}{1000}$th part of a gramme); the English measurement 16·9 minims is equivalent to 1 c.c. (1 gramme) of water at 4° C.

Albumen

The relative quantity of albumen in any specimen of urine may be roughly estimated by the boiling test, already explained. But a more accurate test is to take a measured quantity of urine and allow it to drop into boiling water acidulated with acetic acid. The resulting precipitate is collected on a filter of known weight, washed with boiling water, dried at 212° F., and weighed. The weight of the filter being deducted, the difference will be the weight of albumen in the amount of urine used.

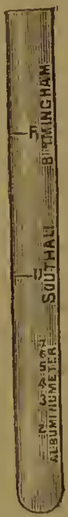

Fig. 15.—Esbach's Albuminometer.

Perhaps, however, the most convenient and accurate method of estimation is by the use of Esbach's albuminometer, which consists of a long and narrow graduated test-tube fixed on a stand. Urine is poured into the tube up to the mark U; the reagent (10 grammes of picric acid and 20 grammes of citric acid in a litre of distilled water) is added up to the mark R. The tube is then closed by the thumb, and the liquids are mixed by reversing it to and fro; shaking should be avoided. The tube is then allowed to stand upright for twenty-four hours, and the height of the coagulum read off, taking the centre of the coagulum as the level. The figure at which the level stands indicates the amount of albumen in grammes per 1,000 c.c., this weight and volume being easily converted into our system thus :

1 gramme (the weight of 1 c.c. aq. dest.) per 1,000 c.c.
=1 c.c. per 1,000 c.c.
=1 part per 1,000 parts
(or 1 in a thousand).

If, for example, the figure on the albuminometer is 4, that is 4 grammes per 1,000 c.c., 4 parts in 1,000, or 0·4 per cent.

When the albumen is very abundant, the urine may be diluted with an equal quantity of water and the result multiplied by 2. If the urine should be neutral or alkaline, it ought to be, as already mentioned, carefully acidified with acetic acid.

Sugar

Estimation by Fehling's reduction test.—Take 10 c.c. of urine and dilute it to the volume of 100 c.c. with distilled water. Pour into a burette, which place over a flask having a capacity of 150 c.c., underneath which is a spirit lamp; now put 10 c.c. of the mixed Fehling's solution and 50 c.c. of distilled water in the flask, and when the contents have commenced to boil let the urine run slowly from the burette into the flask until the blue colour is discharged from the test solution. The level of the liquid in the burette should be noted before starting, and a second reading made when the blue colour is discharged. The boiling of the test solution should be continued until the operation is completed. The number of c.c. of urine mixture used will contain ·05 gramme of grape sugar, and then $\frac{100 \times \cdot05}{\text{c.c. used}}$ is the amount of sugar in the 10 c.c. of urine taken. Thus, supposing it took 50 c.c. of the diluted urine, then a proportion sum arises : if 50 c.c. show the presence of ·05 gramme of sugar, how much will 100 c.c. show? Therefore the sum can be stated :

$$50 : 100 :: \cdot05 : x.$$

As 10 c.c. of urine were taken, the answer refers to this, for there was no grape sugar in the distilled water that diluted it.

A more expeditious way to perform this test is with a Berlin porcelain dish (instead of a flask) and a retort stand, with the burette adjusted to the stem of the retort stand above the dish (see fig. 17).

In estimating with Pavy's solution the method is much the same as with Fehling's solution. The principle is the same, viz. the reduction of the copper to the suboxide, and the discharge of the blue colour. With Pavy's solution, however, there is only the discharge of colour to be looked to, as the ammonia in his solution keeps the suboxide of copper in solution, and 10 c.c. of the solution are equivalent to ·005 glucose. In reading off the number of c.c. of saccharine solution used, the calculation, so as to get a result similar to Fehling's, should read thus :

> As the number of c.c. used : to the total volume of diluted urine :: ·005 : x

—the answer being, as with Fehling's solution, the amount of sugar in the quantity of urine taken before it was diluted with water.

Pavy's process is conducted as follows : 10 c.c. of the ammoniacal cupric solution (equivalent to ·005 gramme of glucose) are diluted with 20 c.c. of distilled water and placed in a small flask ; this is attached by means of a cork to the nozzle of a burette, fitted with a glass stopcock, previously filled with the saccharine urine diluted to a fixed volume. The cork of the flask should also have a small bent tube, to permit steam to escape. The flask is now heated by means of a spirit lamp until the liquid boils, and then the stopcock is turned to allow the saccharine solution to flow into the solution (which should be kept at boiling-point) at the rate of about 100 drops a minute at the beginning ; but when the blue tint nearly fades, the number per minute should be gradually lessened, so as to secure accuracy in exactly discharging the blue tint ; and when this is done, the number of c.c. used is read off and the calculation made. To express parts per 1,000 (a system adopted on the Continent and becoming customary here), suppose 4 c.c. were the amount of diluted urine required to decolorise 10 c.c. of Pavy's solution, then

$$\text{As } 4 : \cdot005 :: 1{,}000 : 1\cdot250, \text{ the answer.}$$

$$\text{Or, } \frac{\cdot005 \times 1{,}000}{4} \text{ or } \frac{5}{4} = 1\cdot250, \text{ the answer.}$$

Another apparatus for the estimation of sugar is Gerrard's glycosometer. Gerrard's method is as follows: Take 5 c.c. of urine containing sugar, and dilute with water to 100 c.c. Now fill two burettes to the zero lines with the diluted urine; next put 10 c.c. of Fehling's solution and 40 c.c. of distilled water into a porcelain dish, and boil. Whilst boiling run the urine first from one burette into the dish in a slow

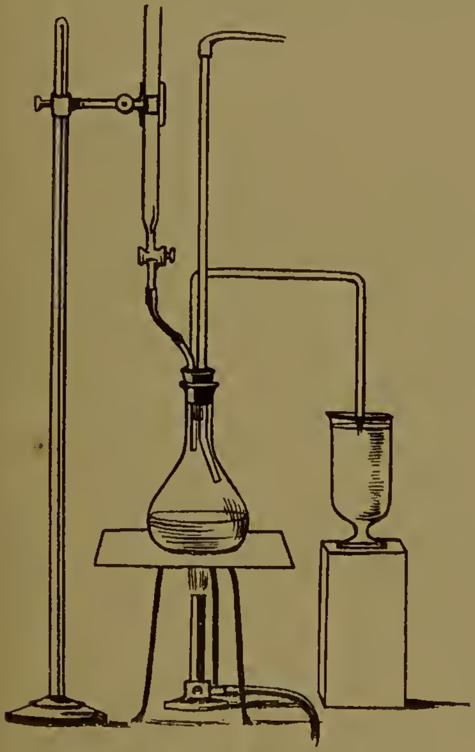

FIG. 16.—ARRANGEMENT OF APPARATUS FOR PAVY'S METHOD (*after Allen*).

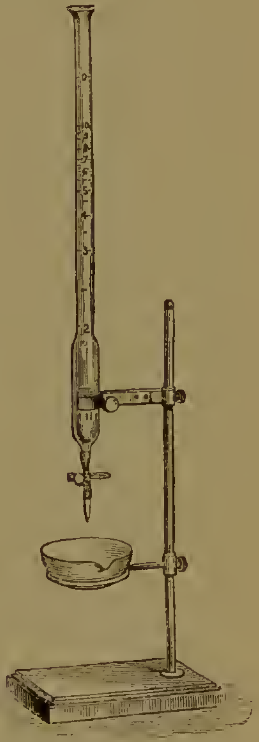

FIG. 17.—APPARATUS FOR GERRARD'S METHOD.

stream, until the blue colour disappears from the Fehling's solution. If the contents of the first burette do not remove the blue colour, continue to use the contents of the second until the colour has gone, when the level of the urine left in the burette shows the percentage of sugar present, and the equivalent in grains per ounce is found by referring to the table sold with the apparatus. The apparatus is graduated to read percentages of sugar between 1 and 10; but if the urine contains more than 10 per cent., 5 c.c. should be diluted with distilled water to 200 c.c. and the result multiplied by 2. [The improved form of burette, fig. 17, is two in one.]

Estimation of Urea

The process known as Hüfner's is based upon the fact that when urea is acted upon by an alkaline hypobromite, nitrogen is given off; and this nitrogen, when collected and measured, gives an estimation of the quantity of urea decomposed. Many forms of apparatus have been contrived to carry out this test. The most recent, and certainly one of the best, is Southall's apparatus. It consists of a bent graduated tube closed at one end, with a funnel-shaped expansion at the other. It is supported on a wooden stand in an inverted position, so that the gas may be collected in the graduated tube, while the liquid displaced by the gas runs into the expanded portion. The hypobromite solution is made by adding bromine to a solution of caustic soda. It must be freshly made as it does not keep long. The solution is made as follows: Sodium hydrate, 5 parts; distilled water, 20 parts; bromine, 2 fluid parts. This will keep for about a

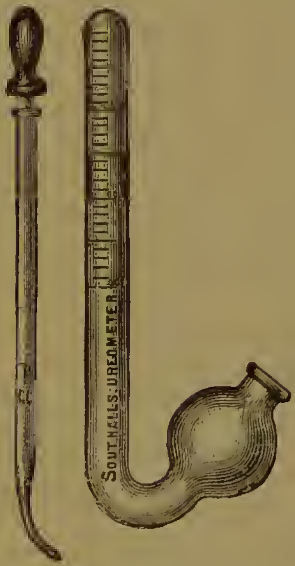

Fig. 18.—Southall's Ureometer.

month. It is convenient to keep the sodium hydrate solution ready, and, when required, to add the bromine in a sealed glass capsule which will break with a shake. In this way the odour of the bromine is avoided. There must be enough hypobromite present to decompose the whole of the urea, and enough sodium hydrate to absorb all the CO_2 produced by the reaction, which is as follows :

$$CON_2H_4 + 3NaBrO = CO_2 + N_2 + 2H_2O + 3NaBr.$$

The directions are :

> Fill the vertical tube with a solution of hypobromite of sodium by pouring the solution into the bulb until it is half full ; incline the apparatus horizontally until the entire tube is filled and just a little left in the bulb (about one-third). Then turn the apparatus to the vertical position, when the fluid in the vertical tube is kept there by atmospheric pressure. Having drawn into the pipette 1 c.c. of the mixed twenty-four hours' collection of urine to be tested, pass the pipette into the apparatus with the point under the long tube, compress the india-rubber cap slowly so that the urine all passes up the long tube. As the urine passes through the solution the urea becomes decomposed and the nitrogen is set free, the gas collecting in the upper part of the tube, where in about fifteen minutes, the froth and bubbles of gas having subsided, its volume, having been measured, indicates the amount of urea from which it is evolved. In cases where there is much urea it is well to test with equal quantities of urine and distilled water, and, when reading the scale, reckon the urea as double that indicated.

The graduations are so arranged that a calculation is unnecessary, the apparatus giving two readings : (*a*) The metric scale, in which each division indicates ·001 gramme of urea in 1 c.c. of urine, the percentage being obtained by multiplying the result by 100. To ascertain the total amount of urea in twenty-four hours, multiply the result of the test by the number of c.c. of urine passed during that time. (*b*) The English scale, in which each division indicates 1 grain of urea per fluid ounce of urine, and the result, multiplied by the number of ounces passed in twenty-four hours, will give the total amount of urea excreted during that period.

Gerrard's ureometer.—Pour into a test-tube 5 c.c. of urine, and into the bottle 25 c.c. of sodium hypobromite solution ; place the test-tube inside the bottle, taking care not to spill any of the contents. Fill the graduated tube with water so that the level reaches the zero line. This is done by removing the clip at the top and pouring water into the side tube, which should be placed high, so that little water remains in it. Now connect the indiarubber tubing to the bottle, and note that the water is at zero. Close the clip. Upset the contents of the test-tube into the hypobromite solution; nitrogen is evolved, and pushes back the water into the side tube. When this ceases, lower the side tube until the levels of the water in both tubes are equal; then note the volume of gas, which shows the percentage of urea.

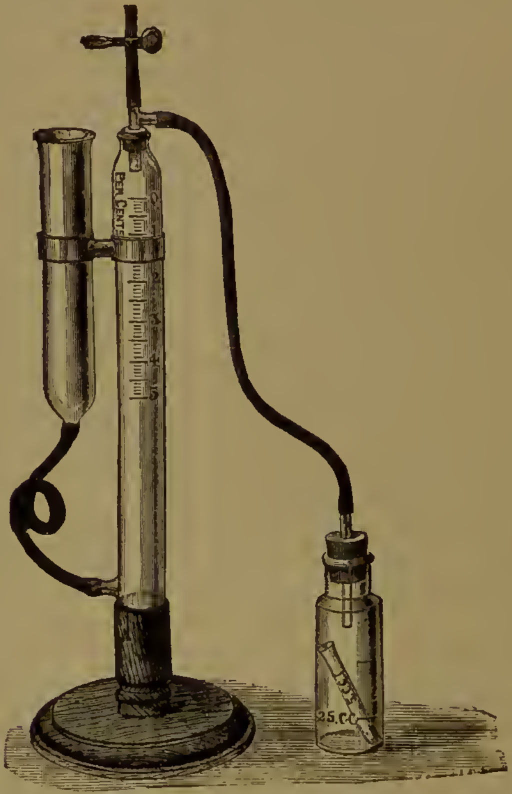

FIG. 19.—GERRARD'S UREOMETER.

The solution of hypobromite of soda is made by dissolving 100 grammes of caustic soda in 250 c.c. of water, and adding 22 c.c of bromine.

To avoid the danger of bromine vapour, the bromine is sold in hermetically sealed glass tubes containing 2·2 c.c. One of these, placed in the large bottle with 25 c.c. of the soda solution, gives, when broken with a sharp shake, the exact quantity of hypobromite for one estimation of urea.

Excess or deficiency test.—Take from the freshly mixed urine of twenty-four hours two portions, one of 5 c.c. and the other of 10 c.c., and place them in watch-glasses. To the smaller portion add one-third its volume of pure colourless nitric acid. If crystals of nitrate of urea appear within a few minutes, the amount of urea is above the normal. Evaporate the other portion to one half its bulk, allow it to cool, and add nitric acid as before. If crystals do not form within a few minutes, the proportion of urea is below the normal.

The quantity of urine passed in twenty-four hours must be taken into consideration. Thus, if instead of the normal quantity the patient passes only half that amount, the first portion should be diluted with its own volume of water, and the second tested without evaporation. If, on the other hand, the urine be that of a diabetic patient, passing perhaps 3,600 c.c. per diem, 15 c.c. of the urine reduced to 5 c.c. by evaporation should be used for the first test, and 30 c.c. reduced to 5 c.c. for the second.

Uric Acid

The estimation of uric acid may be performed by placing 200 c.c. of urine in a beaker; to this must be added 5 c.c. of pure hydrochloric acid. The mixture should be stirred, covered, and set aside for twenty-four hours. The crystals formed are to be collected upon a small weighed filter, washed with cold distilled water until the washings cease to give a precipitate with nitrate of silver, the filter and adhering crystals dried, and weighed. The difference between the total weight and that of the filter will give the amount of uric acid. If the wash-water used does not exceed 30 c.c., no correction is required; but if it does, add to the result ·045 milligramme for each c.c. more than 30 used.

Another method of estimating uric acid consists in mixing the urine with one-twentieth of its bulk of hydrochloric acid and setting it aside for twenty-four hours; collect the deposit on a filter, and wash; then boil it with peroxide of lead and a little water, so as to convert the uric acid into carbonic acid, oxalic acid, allantoin, and urea. The amount of nitrogen in this solution can be estimated by the hypobromite method with a ureometer.

Uric acid contains one-third its weight of nitrogen,[1] so that by multiplying the weight of nitrogen evolved by 3 the quantity of uric acid is obtained, and add ·0045 gramme for each 100 c.c. urine used, to make good loss of uric acid by washing.

Estimation of Phosphates

The solutions required [see p. 55] are:

A standard solution of uranium nitrate, of which 50 c.c. corresponds exactly to 50 c.c. of standard phosphate solution, each c.c. of uranium solution representing ·01 gramme of P_2O_5.

To a solution containing 100 grammes of acetate of sodium dissolved in water, 100 c.c. acetic acid is added, and the mixture made up to 1,000 c.c.

A weak solution of ferrocyanide of potassium.

Of the filtered urine 50 c.c. are mixed in a beaker with 5 c.c. of the acetate solution, the beaker is put on a water bath, and when the mixture is well warmed add uranium solution until the mixture ceases to give a precipitate, which can be ascertained by allowing the solution to trickle down the side of the beaker. A drop from the beaker is placed on a porcelain dish with a glass rod, and a drop of ferrocyanide solution placed near it with another glass rod, and the two drops are allowed to run together. If no alteration of colour be produced at their union, more uranium solution must be added to the urine, until the drops, when meeting, produce a light brown colour. The number of c.c. of uranium solution used is read off, and if after a second warming of the urine in the beaker, and a second trial of the fluid with the ferrocyanide, no increase in the intensity of the colour be produced, the process may be regarded as complete; but if a dark brown colour be formed at the second trial, too much uranium solution has been added, and the estimation must be started again with another 50 c.c. of urine.

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[1] From the volume of nitrogen evolved, the weight of uric acid can be calculated, as 23·55 c.c. of the gas at normal temperature and pressure = 0·084 gramme of uric acid.

To calculate the amount of P_2O_5 passed in twenty-four hours: If 1 c.c. of uranium solution represents ·01 gramme of P_2O_5, and if 15 c.c. of uranium solution have been used, 50 c.c. of urine will contain $\cdot 01 \times 15 = x$ gramme of P_2O_5; and if the patient has passed 1,500 c.c. of urine in twenty-four hours, then the calculation will be $50 : 1{,}500 :: 15 \times \cdot 01 : x$ grammes.

Estimation of Chlorides

Two solutions [see p. 56] are required, viz.:

A standard solution of nitrate of silver, of which 1 c.c. corresponds exactly to 10 milligrammes of chloride of sodium.

A saturated solution of neutral chromate of potash.

The urine should be as fresh as possible; 10 c.c. of the non-albuminous filtered urine are put into a beaker, and two or three drops of the chromate solution added. A burette divided into quarters of a c.c. is filled with the standard solution of nitrate of silver, which solution is allowed to fall into the beaker drop by drop, and is kept well mixed by a glass rod. As each drop of silver solution falls in, it must be closely watched for the least tinge of red surrounding the precipitate of chloride of silver as the termination of the process approaches. But for some time this red is not persistent, disappearing on stirring. The first drop which produces a reddish tinge, not disappearing on stirring, indicates that the whole of the chloride present has been precipitated, and that chromate of silver is about to be formed. The quantity of silver solution is now read off, and another drop let fall into the beaker from the burette, when the reddish colour ought to be stronger. From the number of c.c. used, the quantity of chloride is calculated. Therefore, if 1 c.c. of silver solution corresponds to 10 milligrammes of chloride of sodium, the number of c.c. used of the silver solution multiplied by 10 gives the amount in milligrammes of chloride of sodium present in 10 c.c. of urine. Thus, if 6 c.c. silver solution were used before a permanent red colour was produced, 10 c.c. of urine will contain 60 milligrammes of chloride of sodium; and if the patient passed 1,200 c.c. of urine in twenty-four hours, the amount of chlorides may be calculated thus:

> 10 : 1,200 :: 60 : 7·2 milligrammes = 7·2 grammes, the total quantity passed in the twenty-four hours.

This process always gives a result just a little above the real amount present, so that, if great accuracy is required, 10 c.c. of the urine are measured off into a porcelain crucible, 1 or 2 grammes of chemically pure nitrate of potash are dissolved in it, the whole evaporated to dryness, and exposed to a low red heat until the carbon is completely oxidised and the contents of the crucible are white. After cooling, the residue is dissolved in distilled water and a few drops of nitric acid added until an acid reaction is produced; sufficient precipitated chalk is now added to neutralise the solution, which is then filtered and estimated as in the beginning.

Estimation of Sulphates

For this is required a standard solution of barium chloride, each c.c. of which represents ·04 gramme SO_3 [see p. 56]. Acidulate 50 c.c. of urine with hydrochloric acid, and warm; then add standard solution of barium chloride gradually until the urine ceases to give a precipitate. Now read off the number of c.c. of standard solution used, which multiplied by ·04 will give the amount in grammes of SO_3 in the 50 c.c. of urine taken.

Polariscopic Examination

There is a class of crystals which do not belong to the regular system, but which have the power of double refraction; that is, when a ray of light falls upon any of them it is divided into two, one taking the ordinary course of a refracted ray, the other going in a different direction. Light so altered is said to be polarised, and the two rays are distinguished as the ordinary and the extraordinary ray. There are many ways in which light may be polarised, but one of the most efficient is by a Nicol's prism, which is made by splitting a rhomb of Iceland spar along a diagonal plane and cementing the two parts together by Canada balsam, when one piece will produce polarisation (the polariser) and the other show it (the analyser), and every piece that serves for one of these purposes will also serve for the other. When a ray of light falls upon the first portion of the prism it is polarised—one ray, the ordinary ray, being totally reflected on meeting the Canada balsam and passing out of the prism on the other side; while the extraordinary ray, passing through the balsam and the second portion of the prism, finally emerges at the opposite end to that at which it entered.

Examined by another Nicol's prism, it will be seen that when the two prisms are placed with the chief sections parallel to each other, the ray will pass freely through; but if the analyser be turned round so that it lies at right angles with the polariser, the polarised ray will be entirely reflected by the balsam, therefore the light will not reach the observer's eye. Certain bodies, however, such as quartz, have the power, when placed between the crossed prisms, of giving a colour instead of darkness, due to the twisting of the polarised ray from its original plane, such substances producing what is known as circular polarisation, which may be 'right-handed' (dextro-gyration) or 'left-handed' (lævo-gyration), according as it is necessary to turn the prism to the right or to the left from its proper position to produce again a complete passage of the colourless polarised ray. The rotation by all substances possessing the power of circular polarisation is fixed and increases or diminishes according to the amount of the body present in solution.

There are two varieties of quartz, known as right-handed or left-handed. One rotates the plane of polarisation to the right, the other to the left. When a thin layer of one of these is placed between the two prisms, the ray of polarised light is rotated, and instead of being colourless is coloured, showing all the colours of the spectrum as the analyser is turned, until it once more appears colourless ; and the extent to which the analyser has to be turned (registered by a pointer on the degrees of the circle) is the index of rotary polarisation possessed by the quartz either in a right- or left-handed direction. If the movement of the analyser be continued, colour will again show itself ; but it will be the colour complementary to that first produced. Thus, if in the beginning the colour was red between the uncrossed prisms, and by rotation at an angle of 45° there was no colour, after that the complementary colour would be green, the colour being most intense at an angle of 90°—the prisms being crossed. The polariscope used for the analysis of sugar, and spoken of as a saccharimeter or polarimeter, has the following parts :

1. A Nicol's prism as a polariser.
2. A plate of quartz generally divided down the centre, one side being right- and the other left-handed.
3. A tube to contain the solution to be analysed.
4. A Nicol's prism for rotation, and having a pointer which indicates degrees of the circle on a scale in front.
5. A telescope to focus the line between the two sides of the quartz.

When the pointer is placed at zero, the tube filled with water, and the line focussed, no colour is seen on either side ; but if a solution of sugar be introduced, then a colour is produced on the side of the line corresponding to the nature of the sugar, and then the distance through which the pointer has to be moved round the graduated circle to get both sides of the quartz colourless is the degree of rotary polarisation,[1] the angle through which the prism has been turned is read off from the index and scale attached, and the amount of sugar calculated from this.

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[1] Working with the 100 mm. tube each degree of rotation is equal to 1 per cent. of sugar. With the 200 mm. tube the degrees of rotation divided by 2 give the percentage.

The Spectroscope

When ordinary light undergoes refraction it is broken up into different colours. This is called *chromatic dispersion*, and the colours produced are the spectrum. Sir Isaac Newton first produced a spectrum by cutting a round hole in the shutter of a darkened room, and passing a ray of light through a prism on the inside of the room to a curtain beyond the prism, where it appeared as a long band of different colours. The colours being identical with the colours of the rainbow, this band is spoken of as the solar spectrum. The rainbow is produced when light falls on descending rain, the drops of rain acting as so many prisms. Light therefore is composed of different coloured rays, some of which are bent more than others in passing through the prism. The violet rays are the most bent, and the red the least; the order of colours on the solar spectrum band being—violet, indigo, blue, green, yellow, orange, and red. In most cases the spectra to be observed give such feeble images on screens that an instrument called a spectroscope is employed, the principle of which is similar to that of the hole in the shutter.

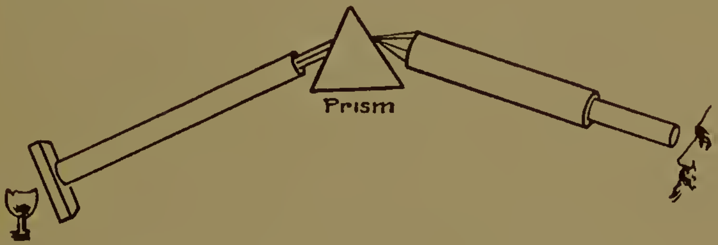

FIG. 20.—DIAGRAM OF THE SPECTROSCOPE.

The spectroscope contains the following parts :

1. A prism to decompose the rays of light into their constituent colours after having passed through the collimator.
2. A brass tube, having at one end a narrow slit in a direction parallel to the edge of the prism, the distance between the parallel edges being

adjustable by a screw, and at the other end of the tube a collimating lens, this being a convex lens with the slit at its principal focus, when the rays from any point of the slit pass through the lens as a parallel pencil, the brass tube being about the focal length of the lens. The tube, lens, and slit are together called the collimator.

3. An observing telescope, generally attached to the prism-stand by a movable joint, and so placed that the rays pass through it after leaving the prism, which must be focussed as if for viewing a distant object.

If, instead of solar light, the light emitted from various bodies in a state of incandescence is directed and condensed by a lens upon a prism, the spectrum produced will in each case be characteristic of the particular bodies employed. It was this discovery which led to the invention of the spectroscope. When solar light is examined by the spectroscope, a number of dark lines are seen to cross the spectrum at fixed points; these are known as 'Fraunhofer's lines,' and are due to the absorption by incandescent elemental vapour of the light which, otherwise, would produce bright bands of colour. Light emitted by a non-volatile body gives a continuous spectrum without lines, and so the light from platinum wire heated in a Bunsen burner in such a position that the rays will pass readily into the spectroscope will not produce visible lines; but if the wire be dipped in hydrochloric acid and a little sodium chloride be taken up and ignited, a bright yellow line will appear in the yellow of the spectrum, so brilliant that it makes all the other yellow part of the image almost invisible. Potassium gives one bright line in the red and one in the blue; lithium, one bright line in the yellow and one in the red; strontium, one blue, one orange, and six red lines. The telescope is arranged to work upon a graduated scale, by which its position for viewing any particular line observed can be noted. The spectroscope therefore is, for substances which can be volatilised, the most delicate test that can be employed. If the proper line is seen, then the constituent searched for is present; if not, it is absent. Fraunhofer described about 600 lines in the solar spectrum, and put a number of them in their proper places in a map of spectra he constructed. The principal lines he distinguished by letters of the alphabet, beginning with A in the red and going on to H in the violet. These lines always occur in the same place as regards colour, but are not at equal distances from one another when prisms of different materials are employed, different parts of the spectrum being unequally expanded by different refracting substances; but this inequality does not prevent identification of the lines.

The spectra of the moon and planets give the dark lines, because they shine by the reflected light of the sun; the fixed stars also give dark lines, but they are different for different stars. If we examine ordinary light which has passed through solutions of various coloured bodies, we get dark lines which are analogous to Fraunhofer's lines, called absorption bands, and which are characteristic of the bodies examined. As a solution of blood shows characteristic absorption bands in the green of the spectrum, the spectroscope is of great service in the examination of urine for blood. A great advantage of this optical process is that it admits of the examination of blood without interfering with chemical or microscopical tests. If the red liquid owes its colour to blood, two dark lines (absorption bands) will be seen, one near the junction of the yellow with the green rays, the other in the middle of the green rays. If the blood is recent and of a red colour, they are distinct and well defined—when viewed by a good artificial or solar light. The spectrum apparatus being properly adjusted, the liquid for examination—being free from sediment—should be as deep in colour as possible. As the spectroscope eye-piece allows of two tubes being examined at once, it is well to compare the suspected liquid with an actual specimen of blood. In the hands of a person skilled in spectroscopic observations, the minutest trace of blood can readily be detected.

It has been stated by an expert in spectrum analysis that the one-thousandth part of a grain of the colouring matter of blood is sufficient to give conclusive evidence of its presence.

Foreign Matters

Extraneous bodies—such as hair, fibres of cotton or wood, fragments of feathers, sputa fæces etc.—are liable to be found in urine, being either purposely placed there for deception or having accidentally fallen in. Therefore in the examination of urine this matter should always be taken into consideration, as many ludicrous mistakes have been made by analysts not on their guard in respect to these casual admixtures. A patient who passes or pretends to pass some extraordinary constituent inconsistent with physical and other diagnostic circumstances should be asked to pass urine either in the presence of the medical attendant or in the presence of some other reliable person. A complete analysis of urine is conveniently tabulated as follows :

REPORT ON SAMPLE OF URINE.

			Normal	Sample
Received from				
Address				
Particulars of collection				
Particulars of quantity				
Physical characters—				
Translucency			Transparent	
Consistency			Limpid	
Colour			Light golden	
Odour			Aromatic	
Froth			Temporary	
Reaction			slightly acid	
Sp. Gr.			1.015 to 1.025	
Total Solids			800 to 1,000 gr. per diem	
Chemical	Normal Constituents	Chlorides	5 to 10 parts per 1,000, as NaCl	
		Sulphates	1·5 to 3 parts per 1,000, as SO_3	
		Phosphates	2 to 3 parts per 1,000, as P_2O_5	
		Urea	2 to 3 per cent.	
		Uric acid	3 to 7 parts per 1,000	
Chemical	Abnormal Constituents	Albumen		
		Albumoses		
		Sugar		
		Bile		
		Bile pigments		
		Bile acids		
		Hæmoglobin		
Microscopical		Oxalates		
		Uric acid		
		Phosphates		
		Cystine		
		Leucine and Tyrosin		
		Cells (pus, blood, etc.)		
		Casts		
		Micro-organisms, as micrococci, bacteria, fungi, sarcinæ, spermatozoa		

Test Reagents

Acid, acetic
Acid, citric
Acid, hydrochloric
Acid, nitric
Acid, sulphuric
Bromine
Liq. ammon. fort.
Liq. potassæ
Mercurous nitrate
Methyl violet
Ozonic ether
Plaster of Paris
Plumbi acetas
Plumbi peroxidum
Potassii hydras
Soda tartarata
Sodii hydras
Sol. arg. nit. (10 gr. ad ℥j)
Sol. cupri sulph. (10 gr. ad ℥j)
Sol. ferric chloride (neutral)
Sp. vini rect.
Tr. guaiaci
Tr. iodi
Yeast (compressed)

Magnesia mixture—Dissolve magnes. carb. in a slight excess of hydrochloric acid, add one-third its bulk of liq. ammon. fort., then stir in ammonium chloride until the precipitate is dissolved.

Standard Test Solutions

Fehling's Solution

This solution is made and kept in two parts, and, when required for use, equal volumes are mixed. On heating the mixed liquids in a test-tube to 212° F. the mixture should remain quite clear. Each c.c. of the mixed liquid represents ·05 gramme of glucose.

No. 1

Sulphate of copper 346·4 grains
Distilled water to make 5,000 grain measures

No. 2

Caustic soda 1¾ ounce
Tartarated soda 4 ounces
Distilled water to make 5,000 grain measures

Pavy's Solution

Dissolve 20·4 grammes of Rochelle salt and the same weight of caustic potash in distilled water. Dissolve separately 4·158 grammes of cupric sulphate in more water, with heat; add this to the solution first prepared; when cold, add 300 c.c. of strong ammonia, and then make up with distilled water to a litre. Ten c.c. of this ammoniated cupric solution should have its colour exactly discharged by a portion of urine containing ·005 gramme of glucose.

Standard Solutions for Chlorides, Phosphates, and Sulphates

1. *Uranium solution.*—Dissolve 70 grammes of uranic nitrate in 900 c.c. of water, and then ascertain its strength by making an analysis

of 50 c.c. of the standard phosphate solution, diluting with water so that 50 c.c. will correspond exactly to 50 c.c. of that solution and balance the standard phosphate solution, each 1 c.c. being equal to ·01 gramme P_2O_5.

2. *Standard phosphate solution.*—Dissolve 50·42 grammes of pure di-sodium hydrogen phosphate in 900 c.c. of water, and make up to a litre with water, when each 1 c.c. will equal ·01 gramme of P_2O_5.

3. *Sodium acetate solution.*—A solution of 100 grammes of sodium acetate and 100 grammes of acetic acid made up to a litre with water.

4. A weak yellowish solution of ferrocyanide of potassium.

5. *Standard solution of $AgNO_3$ for the estimation of chlorides.*—Decinormal solution of pure nitrate of silver, in practice 17 grammes (in theory 16·966) dissolved in a litre of distilled water. 1 c.c. = ·00585 gramme NaCl. If normal solution be used, shift the decimal one place to the left.

6. *Standard solution of $BaCl_2$ for the estimation of sulphates.*—Normal solution: 121·77 grammes of pure barium chloride dissolved in a litre of distilled water. 1 c.c. = ·04 gramme SO_3.

Apparatus

Albuminometer (Esbach's).
Beakers.
Berlin porcelain dishes.
Filters (paper).
Flask (fitted with bent glass tube in the cork to allow escape of steam; also a hole to receive the nozzle of a burette for use with Pavy's or Fehling's solution).
Funnels (glass).
Glycosometer (Gerrard's).
Litmus paper (red and blue).
Measure (a litre measure divided into at least 10 c.c. parts).
Microscope.
Pipettes (which deliver 5, 10, 15, 20, 30 and 50 c.c.).
Polariscope (for saccharimeter).
Retort-stand.
Rods (glass).
Spectroscope.
Spirit lamp.
Test-tubes.
Test-tube stands.
Test-tube holders.
Ureometer (Southall's or Gerrard's).
Urinometer (Fletcher's).
Urine test-glasses.
Watch-glasses.
Metric weights and measures.

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